BARORECEPTOR-SENSITIVE FLUCTUATIONS OF HEART RATE AND PUPIL DIAMETER

G. Calcagnini, P.Giovannelli, F. Censi, P. Bartolini, V. Barbaro

Biomedical Engineering Laboratory, Istituto Superiore di Sanità, Roma, Italy

Abstract - **It is generally known that the pupil is under the control of the autonomic nervous system. Recently, those rhythms characterizing the autonomic fluctuations of heart period and arterial blood pressure have been detected in spontaneous Pupil Diameter (PD) fluctuations. The physiological mechanisms underlying such variability have not been widely investigated. Aim of this study was to investigate the origin of the pupil fluctuations in humans, using a non-invasive modulation of carotid baroreceptors by Neck Suction (NS). To this purpose, we simultaneously recorded ECG, respiration activity, NS pressure and PD fluctuations from 10 normal subjects. The equipment for the PD measurement and the NS stimulation was developed in our laboratory. The response of the pupil to the NS was studied at stimulation frequencies of 0.10 and 0.20 Hz, by using parametric spectral and cross-spectral estimation. In all subjects, the NS rhythms were clearly detectable in heart rate variability series in both stimulation frequencies and also in the PD spectra with significant coherences (>0.5). These findings suggest that blood pressure fluctuations propagate to the pupil via carotid baroreceptor afferent pathways. However a central contribution can not be excluded.**

Keywords **– pupil diameter fluctuations, heart rate variability, baroreceptors, spectral analysis**

I. INTRODUCTION

The pupil of the human eye continuously fluctuates even in absence of visual accommodation and/or light stimulation. The sympathetic and parasympathetic innervations of the iris muscle are supposed to be responsible for these physiologic fluctuations of the human pupil (hippus). It has been demonstrated, first in animals [1] and then in humans [2][3], that the respiratory rhythm is one of the source of these fluctuations. In general the inspiration is accompanied by pupil dilatation, the expiration by pupil constriction.

The respiratory rhythm, detectable in several sympathetic and parasympathetic nervous fibers and in the Heart Rate Variability (HRV) signal [4], is referred to as the High Frequency (HF) component (0.15-0.4 Hz), and is considered a marker of the parasympathetic control to the sinus node. It is well assessed that in HRV signal a relatively slower component is also detectable, in the so-called Low Frequency (LF) band (0.04-0.15 Hz). This component characterizes the peripheral sympathetic nerves too, and is considered expression of both sympathetic and parasympathetic control. Recently, the identification of a spectral component in the LF band in spontaneous Pupil Diameter (PD) fluctuations has been reported by our group [5] and by others [6]. Animal studies suggested that two sources for pupil oscillations could be involved: central respiratory activity and respiratory blood pressure fluctuations that modulate pupil width via sinoaortic baroreceptors [1]. It might be hypothesized that in humans carotid and sinoaortic baroreceptors may contribute to, if not cause, the genesis of pupillary LF and HF rhythms. A non-invasive investigation of the human baroreceptor function can be performed by the Neck Suction (NS) technique [7][8]. This technique allows a periodic modulation of the carotid sinus baroreceptors, and stimulates the brainstem nuclei through the afferent parasympathetic pathways.

Aim of this study is to investigate the origin of spontaneous pupil fluctuations in humans, using a non-invasive modulation of carotid baroreceptors by NS. In addition, to distinguish between the sympathetic and parasympathetic efferent activity we studied the response of the pupil to the NS at different frequencies of stimulation. The equipment for the PD measurement and for the NS stimulation were fully developed in our laboratory.

II. METHODOLOGY

A. Experimental protocol and data acquisition

5 healthy young subjects (24-28 years) were studied. During the experiments, the lighting condition and the temperature of the laboratory were held constant. NS was carried out and monitored in the supine position during controlled breathing at 15 breaths/min.

The neck suction was applied to two cuffs posed on both sides of the neck (at the positions corresponding to the carotid baroreceptors). The cuffs have been connected to a vacuum source, whose power was modulated by a software-controlled feed-back sinusoidal control (figure 1). The use of the cuffs instead of a molded collar (previously employed [9]) is more comfortable for the patient, although providing the same stimulation.

A pressure transducer was used to measure the air pressure inside the cuffs. The pressure signal was used as the feed-back signal for the control loop. The input signal of the control circuit was provided by the same PC used for the acquisition.

The protocol consisted in two stages lasting 5 minutes each. The NS stimulation frequency was set to 0.2 Hz in the first stage and to 0.1 Hz in the second one. Control breathing condition at rates of 15 breaths/minute (0.25 Hz) was used to have a clear discrimination between LF, HF and NS rhythms. The subjects were instructed initiate a breath with each tone of a series of auditory cues and to look at a target panel 1 meter in front of them. Light intensity of the panel ...

ECG, Respiration Activity (RA), Neck Suction Pressure (NSP) and Pupil Diameter (PD) were simultaneously recorded (figure 1). RA was monitored by a pletismographic thoracic belt. ECG, RA and NSP were continuously recorded and real-time sampled (sampling frequency: 500 Hz, resolution: 12 bit) via an A/D converter board (DAQCard

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Abstract

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1200, National Instruments), plugged into a laptop. We continuously measured the PD fluctuations by a portable, infrared TV-pupillometer. The images of pupil have been captured by a micro infrared CCD camera mounted on a light helmet and connected to a video capture board (PCI 1408, National Instruments) for the real-time digitising of the image sequences. The camera was equipped with an infrared filter to eliminate reflexes from the natural light source in the room.

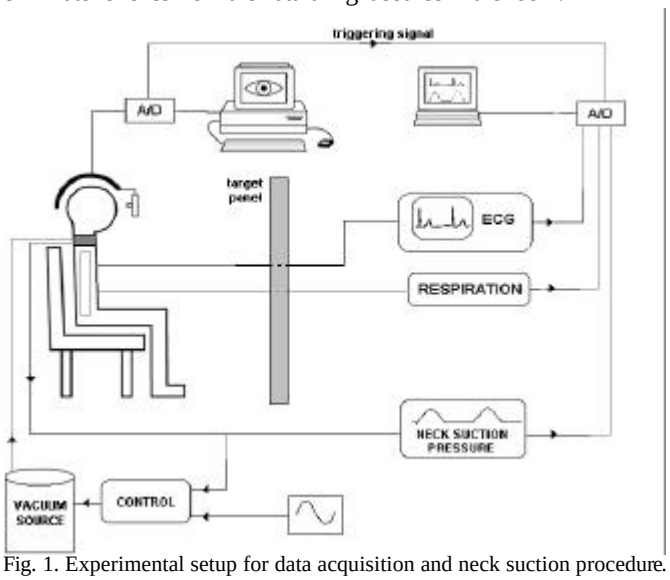

Fig. 1. Experimental setup for data acquisition and neck suction procedure.

The proper illumination was provided by a near infrared diode (780nm). A frame rate of 12.5 frames/s and a resolution of 768x576, 256 grey levels were chosen as a trade off among data storage, computing time, spatial resolution and pupillary dynamic bandwidth constrains. The synchronisation of the 2 acquisition systems was obtained by a triggering signal, the frame synchronisation signal, which provides a maximum delay of 40 ms. Figure 2 shows two images of the equipment for the PD measurements and the cuffs used for the NS stimulation.

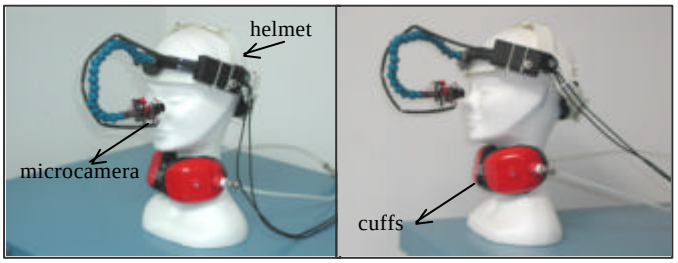

Fig. 2. Equipment for the PD measurements and cuffs used for the NS stimulation.

B. Estimation of pupil diameter

Measurements of the pupil diameter have been performed by a two steps procedure: first, the points laying on the pupil boundary have been detected by using a curvature algorithm [10]; these points have been then interpolated according to the method of Chauduri and Kundu [11], based on an optimum weighted least square circular fitting.

For each acquisition, calibration has been performed by acquiring images of known diameter circumferences located on the subject eyelid (figure 3). The calibration procedure has been performed at the end of each acquisition.

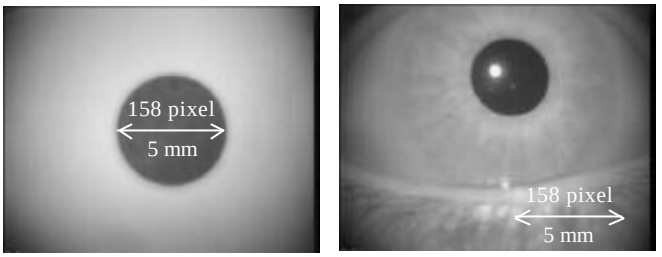

Fig. 3. Calibration procedure: known diameter circumference located on the subject eyelid (left) and relative subject pupil image (right).

C. Correction of blinking artifacts

The algorithm used for the estimation of pupil diameter (curvature algorithm and circular fitting) also detects when the pupil is partially or totally covered by the eyelid (blinking artifacts). In these cases, the pupil diameter is set to 0 mm. Correction of blinking artifacts has been performed by reconstructing these missing data through a cubic spline interpolation.

D. Construction of variability series

Two approaches are generally used to obtain proper representation for cardiovascular variability data: the beat-to-beat approach [12] and the low pass filtering approach [13]. In this study we extract the variability series according to the latter approach, in order to avoid any distortions in PD fluctuations from NS modulation of the heart period.

The low-pass filtered event series (ES) was used to extract the heart rate variability (HRV) signal since it is a reliable time domain representation with high temporal resolution. In ES each beat is replaced by a δ function; the signal can be described as:

$$x(t) = \sum_{k=0}^{N-1} \delta(t - t_k)$$

where δ is the Dirac delta function, t_k is the occurrence time of the k^{th} beat and N is the total number of beats. To obtain the heart rate variability series, the ES was low-pass filtered at 0.5 Hz and re-sampled at 1.25 Hz [13]. Correspondingly, RA, NSP and PD variability series were obtained by a similar low-pass filtering procedure (FIR, 10 order, cut-off frequency 0.5 Hz) and a re-sampling at 1.25 Hz of the RA, NSP and PD series, respectively. According to this procedure, HRV and RA series were expressed in arbitrary units (a.u.), NSP in mmHg and PD in mm. This approach particularly guarantees the time synchronization between HRV, RA, NSP and PD.

III. RESULTS

To detect the effect of the sinusoidal stimulation over the PD and HRV series, we used a parametric spectral and cross-spectral estimation by autoregressive (AR) modelling over 250 consecutive samples.

We analyzed the HRV series to assess the efficacy of the NS stimulation in eliciting a baroreflex response.

With the NS series as a reference, the spectral coherences were used to assess whether NS induced synchronous fluctuations in PD.

Figure 4 shows the results obtained for the 0.2 Hz stimulation, in one subject: time series and monovariate AR spectra are shown. HRV spectrum demonstrates that the NS

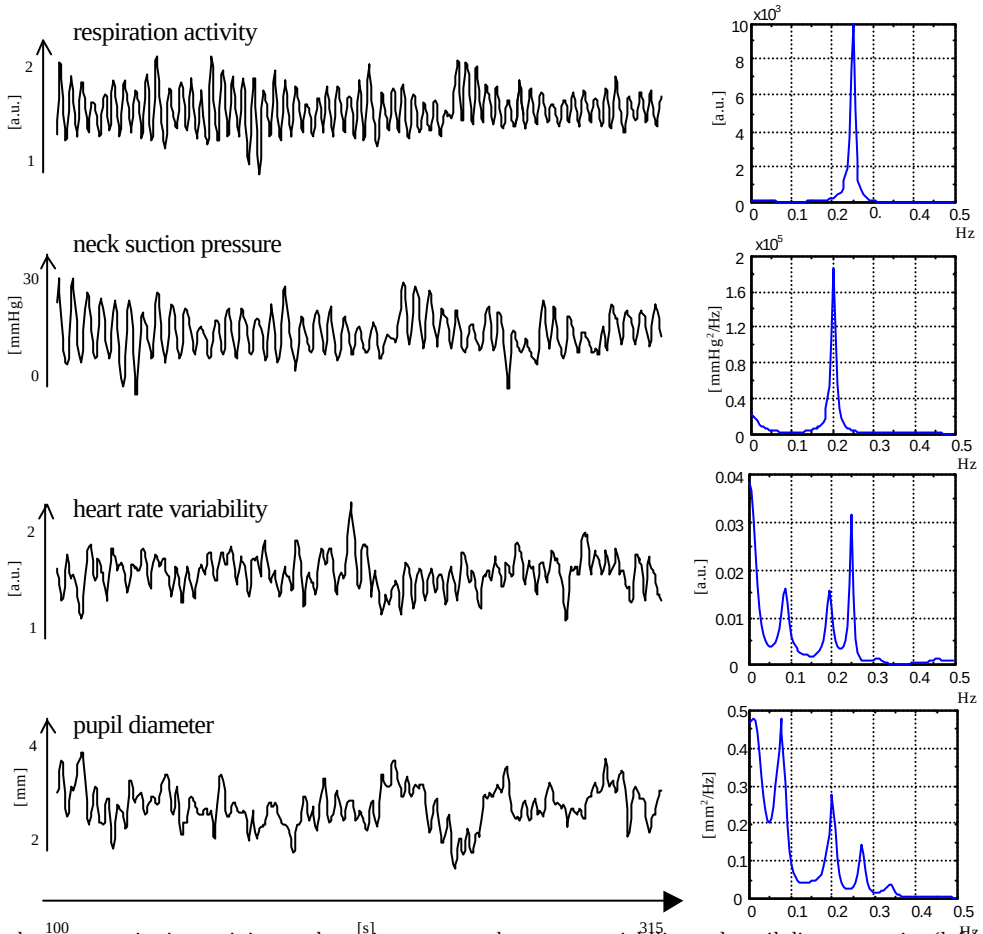

Fig. 4. From top to bottom: respiration activity, neck suction pressure, heart rate variability and pupil diameter series (left) together with the corresponding spectra (right), for one subject.

stimulation elicits the physiological baroreflex response (i.e. slowing of heart rate when carotid are stretched). Controlled respiration provides a respiratory component (0.25 Hz) clearly distinguishable from NS. The PD spectrum reveals three main harmonic components at 0.07 Hz, 0.20 Hz and 0.25 Hz. The 0.20 and 0.25 Hz components correspond to the NS stimulation and the controlled breathing, respectively.

Figure 5, upper panel, reports the spectral coherence between HRV series and PD during the 0.20 Hz NS stimulation, for the same subject as figure 4. Note the significant values reached at 0.20 and 0.25 Hz.

Similar results were obtained for the NS stimulation at 0.1 Hz, as shown in figure 5, lower panel. Baroreflex stimulation by NS was observed in PD spectra in the other 4 subjects, but the relative and absolute amplitude differed.

IV. DISCUSSION

To our knowledge the contribution of baroreflex control to the spontaneous PD fluctuations was only investigated by invasive study in animals [1].

Borgdorff investigated the origin of the pupil fluctuations in cats, using an invasive approach which included artificial ventilation, baroreceptor denervation and direct electrical stimulation of pulmonary vagal afferent fibers.

Borgdorff concluded that at least two sources contribute to the respiratory rhythm in cat PD: the rhythmic activity of the respiratory centre and the respiratory blood pressure fluctuations that modulate pupil width via sinoaortic baroreceptors.

Ohtsuka et al. reported respiratory fluctuations in pupil in 6 normal subjects at different respiration frequencies and tidal volumes [2]. They found that the amplitude of the respiratory fluctuations of the pupil area were closely proportional to the tidal volumes. Their experimental setup did not allow to investigate the mechanisms by which the respiratory rhythm propagates to the pupil.

Our study performed a non-invasive investigation not only of the respiratory pupillary fluctuations in humans, but also of the LF rhythm, which was recently reported by our group and others [5][6].

Our findings showed that a baroreceptor stimulation, miming blood pressure changes in LF and HF bands, caused PD fluctuations at the same frequency. Because the influence of blood pressure on PD can neither be mediated by an increased filling of the iris vessel nor by fluctuations in intraocular pressure [1], a neuro-mediated mechanism can be hypothesised [14]. We can speculate that signals from the carotid sinus reach not only the cardiovascular centre, but almost the entire reticular formation. Here, rhythmic impulses induce fluctuations that are conveyed to various organs,

including the pupils. Whether and to which extent a central activity stemming from the respiratory centre or from the hypothalamus contributes to PD fluctuations, was not addressed by our study.

V. CONCLUSION

Our aim was to investigate whether the spontaneous LF and HF rhythms in pupil fluctuations may result from the LF and HF fluctuations of blood pressure, mediated by the carotid sinus baroreceptors. We found that the carotid baroreceptor stimulation induces pupil fluctuations locked to the stimulation frequency. Thus it can be speculated that blood pressure fluctuations in LF and HF bands contribute to the spontaneous fluctuations of human pupil, via afferent carotid baroreceptor pathways, even if a central contribution cannot be excluded.

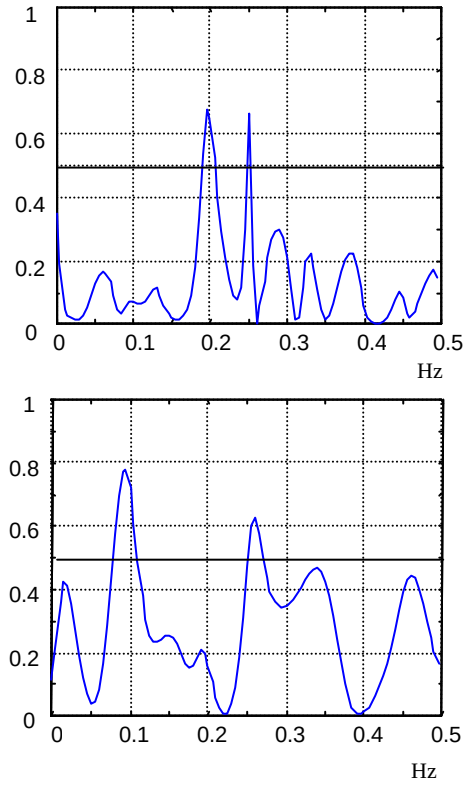

Fig. 5. Spectral coherences between HRV series and PD during the 0.20 Hz (top) and 0.1 Hz (bottom) NS stimulation, for the same subject as figure 4.

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